DIALOG(R)File 155:MEDLINE(R)

09120994 97146676 PMID: 8993532
Signals, receptors, and cytosolic factors involved in **peroxisomal** protein import.
Terlecky SR; Wiemer EA; Nuttley WM; Walton PA; Subramani S
Department of Biology, University of California, San Diego, La Jolla 92093-0322, USA.
Annals of the New York Academy of Sciences (UNITED STATES) Dec 27 **1996**, 804 p11-20, ISSN 0077-8923 Journal Code: 5NM
Languages: ENGLISH
Document type: Journal Article; Review; Review, Academic
Record type: Completed
Peroxisomes are ubiquitous eukaryotic organelles which function in a wide variety of metabolic processes. The many lethal human disorders associated with defects in **peroxisomal** protein import underscore the importance of this organelle. In recent years, the evolutionarily conserved molecular mechanisms of protein **targeting** to, and translocation across, **peroxisomal** membranes have begun to emerge. Signals which route proteins to the organelle have been identified, as have cytosolic, membrane-associated, and lumenal components of the import machinery. The goal of this brief **review** was to summarize our current knowledge of some of these molecules and to describe several potential mechanisms by which **peroxisomes** selectively import their constituent proteins. Aspects of these mechanisms that distinguish **peroxisomal** protein

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Set     Items   Description
S1          0   PEROXISOM$ AND TARGET$ AND SACCHAROMYCES AND PY<1998
S2          0   PEROXISOM$ AND TARGET$ AND SACCHAROMYCES
S3          0   PEROXISOM$ AND SACCHAROMYCES
S4        281   PEROXISOM? AND TARGET? AND SACCHAROMYCES
S5    2580950   4 AND PY<1998
S6        172   S4 AND PY<1998
S7        109   RD (unique items)
S8          4   S7 AND REVIEW
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            109  S7
              4  S8
      S9    105  S7 NOT S8
? t s9/3,ab/1-20


 9/3,AB/1     (Item 1 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09647566   98087270   PMID: 9427398
    Regulation of the yeast SPS19 gene encoding peroxisomal
2,4-dienoyl-CoA reductase by the transcription factors Pip2p and Oaf1p:
beta-oxidation is dispensable for Saccharomyces cerevisiae
sporulation in acetate medium.
  Gurvitz A; Rottensteiner H; Hiltunen JK; Binder M; Dawes IW; Ruis H;
Hamilton B
  School of Biochemistry and Molecular Genetics, University of New South
Wales, Sydney, Australia.
  Molecular microbiology (ENGLAND)   Nov 1997,  26  (4)  p675-85,
ISSN 0950-382X   Journal Code: MOM
  Languages: ENGLISH
  Document type: Journal Article
  Record type: Completed
  The yeast SPS19 gene encoding the peroxisomally targeted
2,4-dienoyl-CoA reductase shares its promoter region (291 bp) with the
sporulation-specific gene SPS18. SPS19 is induced during sporulation in
diploids but to a lesser extent than SPS18; under oleate induction
conditions, SPS19, but not SPS18, is transcribed via an oleate response
element (ORE) independently of ploidy or sporulation. The SPS19 ORE is the
binding target of the Pip2p and Oaf1p transcription factors, and an
SPS19-lacZ reporter gene, which is highly expressed in oleate-induced
cells, is not activated in haploids devoid of either protein. We examined
the expression of CYC1-lacZ reporter constructs carrying the SPS19 and CTA1
OREs in diploids propagated under sporulation conditions and have shown
that OREs are not sufficient for heterologous expression during yeast
development. In addition, diploids deleted at either PIP2 or OAF1
demonstrated abundant ascosporogenesis, indicating that these genes are not
essential for sporulation. A deltapex6 strain lacking peroxisomal
structures and one devoid of fatty acyl-CoA oxidase (deltapox1), the first
step in fungal beta-oxidation, were both proficient for sporulation and,
hence, beta-oxidation and the peroxisomal compartment containing it
are dispensable for meiotic development.


 9/3,AB/2     (Item 2 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
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09645405 98070310 PMID: 9405362

Overexpression of Pex15p, a phosphorylated **peroxisomal** integral membrane protein required for **peroxisome** assembly in S.cerevisiae, causes proliferation of the endoplasmic reticulum membrane.

Elgersma Y; Kwast L; van den Berg M; Snyder WB; Distel B; Subramani S; Tabak HF

Department of Biology, University of California at San Diego, Bonner Hall, 9500 Gilman Drive, La Jolla, CA 92093-0322, USA.

EMBO journal (ENGLAND) Dec 15 **1997**, 16 (24) p7326-41, ISSN 0261-4189 Journal Code: EMB

Contract/Grant No.: DK41737, DK, NIDDK

Languages: ENGLISH

Document type: Journal Article

Record type: Completed

We have cloned PEX15 which is required for **peroxisome** biogenesis in **Saccharomyces** cerevisiae. pex15Delta cells are characterized by the cytosolic accumulation of **peroxisomal** matrix proteins containing a PTS1 or PTS2 import signal, whereas **peroxisomal** membrane proteins are present in **peroxisomal** remnants. PEX15 encodes a phosphorylated, integral **peroxisomal** membrane protein (Pex15p). Using multiple in vivo methods to determine the topology, Pex15p was found to be a tail-anchored type II (Ncyt-Clumen) **peroxisomal** membrane protein with a single transmembrane domain near its carboxy-terminus. Overexpression of Pex15p resulted in impaired **peroxisome** assembly, and caused profound proliferation of the endoplasmic reticulum (ER) membrane. The lumenal carboxy-terminal tail of Pex15p protrudes into the lumen of these ER membranes, as demonstrated by its O-glycosylation. Accumulation in the ER was also observed at an endogenous expression level when Pex15p was fused to the N-terminus of mature invertase. This resulted in core N-glycosylation of the hybrid protein. The lumenal C-terminal tail of Pex15p is essential for **targeting** to the **peroxisomal** membrane. Furthermore, the **peroxisomal** membrane **targeting** signal of Pex15p overlaps with an ER **targeting** signal on this protein. These results indicate that Pex15p may be **targeted** to **peroxisomes** via the ER, or to both organelles.

9/3,AB/3 (Item 3 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09630740 98092307 PMID: 9432010

Gene analysis of an NADP-linked isocitrate dehydrogenase localized in **peroxisomes** of the n-alkane-assimilating yeast Candida tropicalis.

Kawachi H; Shimizu K; Atomi H; Sanuki S; Ueda M; Tanaka A

Department of Synthetic Chemistry and Biological Chemistry, Graduate School of Engineering, Kyoto University, Japan.

European journal of biochemistry (GERMANY) Nov 15 **1997**, 250 (1) p205-11, ISSN 0014-2956 Journal Code: EMZ

Languages: ENGLISH

Document type: Journal Article

Record type: Completed

In n-alkane-utilizing yeast, Candida tropicalis, two NADP-linked isocitrate dehydrogenase (NADP-IDH) isozymes are present, one in mitochondria (Mt-NADP-IDH) and the other in **peroxisomes** (Ps-NADP-IDH). Here we report the isolation, sequencing, and expression of the gene encoding Ps-NADP-IDH (CtIDP2), distinct from the Mt-NADP-IDH gene (CtIDP1). Based on the N-terminal amino acid sequence of purified Ps-NADP-IDH, a cDNA fragment specific for Ps-NADP-IDH was obtained by the 5'-RACE method. Using this fragment as a probe, the genomic CtIDP2 gene was isolated. Nucleotide sequence analysis of CtIDP2 disclosed that the region encoding CtIdp2p had a length of 1233 bp, corresponding to 411 amino acid residues. The deduced N-terminal amino acid sequence matched the results obtained from the purified protein. When this CtIDP2 was expressed in **Saccharomyces** cerevisiae using the C. tropicalis isocitrate lyase

Record type: Completed
Most **peroxisomal** matrix proteins contain a carboxyl-terminal tripeptide that directs them to **peroxisomes**. Within limits, these amino acids may be varied, without loss of function. The specificity of this **peroxisomal targeting** signal (PTS1) is remarkable considering its small size and its relaxed consensus sequence. Moreover, several **peroxisomal** proteins have a PTS1-like signal that does not fit the reported consensus sequence. Because many of these PTS1 variants seem to be functional in a species-dependent or protein context-dependent manner, we investigated the PTS1 requirements in a homologous context, using **Saccharomyces** cerevisiae and endogenous **peroxisomal** malate dehydrogenase (MDH3). **Peroxisomal** import of the MDH3-PTS1 variants was tested qualitatively by the ability to complement the Deltamdh3 mutant and quantitatively by subcellular fractionation. We observed efficient import of MDH3 into **peroxisomes** with a large variety of PTS1 tripeptides. Many of these variants do not fit the observed PTS1 requirements for heterologously expressed proteins, which suggests that additional domains in the protein may be of decisive importance whether or not a certain PTS1 variant is recognized by the components of the **peroxisomal** import machinery. Because we show that dimerization of MDH3 precedes import into the organelle, these domains are most likely conformational domains.

9/3,AB/6 (Item 6 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09516119 96354904 PMID: 8769411
Targeting of human catalase to **peroxisomes** is dependent upon a novel COOH-terminal **peroxisomal targeting** sequence.
Purdue PE; Lazarow PB
Department of Cell Biology and Anatomy, Mount Sinai School of Medicine, New York, New York 10029, USA.
Journal of cell biology (UNITED STATES) Aug **1996**, 134 (4) p849-62, ISSN 0021-9525 Journal Code: HMV
Contract/Grant No.: DK19394, DK, NIDDK
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
We have identified a novel **peroxisomal targeting** sequence (PTS) at the extreme COOH terminus of human catalase. The last four amino acids of this protein (-KANL) are necessary and sufficient to effect **targeting** to **peroxisomes** in both human fibroblasts and **Saccharomyces** cerevisiae, when appended to the COOH terminus of the reporter protein, chloramphenicol acetyl transferase. However, this PTS differs from the extensive family of COOH-terminal PTS tripeptides collectively termed PTS1 in two major aspects. First, the presence of the uncharged amino acid, asparagine, at the penultimate residue of the human catalase PTS is highly unusual, in that a basic residue at this position has been previously found to be a common and critical feature of PTS1 signals. Nonetheless, this asparagine residue appears to constitute an important component of the catalase PTS, in that replacement with aspartate abolished **peroxisomal targeting** (as did deletion of the COOH-terminal four residues). Second, the human catalase PTS comprises more than the COOH-terminal three amino acids, in that COOH-terminal-ANL cannot functionally replace the PTS1 signal-SKL in **targeting** a chloramphenicol acetyl transferase fusion protein to **peroxisomes**. The critical nature of the fourth residue from the COOH terminus of the catalase PTS (lysine) is emphasized by the fact that substitution of this residue with a variety of other amino acids abolished or reduced **peroxisomal targeting**. **Targeting** was not reduced when this lysine was replaced with arginine, suggesting that a basic amino acid at this position is required for maximal functional activity of this PTS. In spite of these unusual features, human catalase is sorted by the PTS1 pathway, both in yeast and human cells. Disruption of the PAS10 gene

encoding the S. cerevisiae PTS1 receptor resulted in a cytosolic location of chloramphenicol acetyl transferase appended with the human catalase PTS, as did expression of this protein in cells from a neonatal adrenoleukodystrophy patient specifically defective in PTS1 import. Furthermore, through the use of the two-hybrid system, it was demonstrated that both the PAS10 gene product (Pas10p) and the human PTS1 receptor can interact with the COOH-terminal region of human catalase, but that this interaction is abolished by substitutions at the penultimate residue (asparagine-to- aspartate) and at the fourth residue from the COOH terminus (lysine-to-glycine) which abolish PTS functionality. We have found no evidence of additional **targeting** information elsewhere in the human catalase protein. An internal tripeptide (-SHL-, which conforms to the mammalian PTS1 consensus) located nine to eleven residues from the COOH terminus has been excluded as a functional PTS. Additionally, in contrast to the situation for S. cerevisiae catalase A, which contains an internal PTS in addition to a COOH-terminal PTS1, human catalase lacks such a redundant PTS, as evidenced by the exclusive cytosolic location of human catalase mutated in the COOH-terminal PTS. Consistent with this species difference, fusions between catalase A and human catalase which include the catalase A internal PTS are **targeted**, at least in part, to **peroxisomes** regardless of whether the COOH-terminal human catalase PTS is intact.

9/3,AB/7 (Item 7 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09515917 96324986 PMID: 8702562

Isolation and characterization of Pas2p, a **peroxisomal** membrane protein essential for **peroxisome** biogenesis in the methylotrophic yeast Pichia pastoris.

Wiemer EA; Luers GH; Faber KN; Wenzel T; Veenhuis M; Subramani S
Department of Biology, University of California at San Diego, La Jolla, California 92093-0322, USA.

Journal of biological chemistry (UNITED STATES) Aug 2 **1996**, 271 (31) p18973-80, ISSN 0021-9258 Journal Code: HIV
Contract/Grant No.: NIHDK41737, HD, NICHD
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

The pas2 mutant of the methylotrophic yeast Pichia pastoris is characterized by a deficiency in **peroxisome** biogenesis. We have cloned the PpPAS2 gene by functional complementation and show that it encodes a protein of 455 amino acids with a molecular mass of 52 kDa. In a Pppas2 null mutant, import of both **peroxisomal targeting** signal 1 (PTS1)- and PTS2-containing proteins is impaired as shown by biochemical fractionation and fluorescence microscopy. No morphologically distinguishable **peroxisomal** structures could be detected by electron microscopy in Pppas2 null cells induced on methanol and oleate, suggesting that PpPas2p is involved in the early stages of **peroxisome** biogenesis. PpPas2p is a **peroxisomal** membrane protein (PMP) and is resistant to extraction by 1 M NaCl or alkaline sodium carbonate, suggesting that it is a **peroxisomal** integral membrane protein. Two hydrophobic domains can be distinguished which may be involved in anchoring PpPas2p to the **peroxisomal** membrane. PpPas2p is homologous to the **Saccharomyces** cerevisiae Pas3p. The first 40 amino acids of PpPas2p, devoid of the hydrophobic domains, are sufficient to **target** a soluble fluorescent reporter protein to the **peroxisomal** membrane, with which it associates tightly. A comparison with the membrane **peroxisomal targeting** signal of PMP47 of Candida boidinii revealed a stretch of positively charged amino acids common to both sequences. The role of **peroxisomal** membrane **targeting** signals and transmembrane domains in anchoring PMPs to the **peroxisomal** membrane is discussed.

9/3,AB/8 (Item 8 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09500144 95386582 PMID: 7544797
Identification of three distinct **peroxisomal** protein import defects in patients with **peroxisome** biogenesis disorders.
Slawecki ML; Dodt G; Steinberg S; Moser AB; Moser HW; Gould SJ
Kennedy Krieger Institute, Baltimore, MD 21205, USA.
Journal of cell science (ENGLAND) May **1995**, 108 (Pt 5) p1817-29, ISSN 0021-9533 Journal Code: HNK
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Zellweger syndrome, neonatal adrenoleukodystrophy, infantile Refsum's disease, and classical rhizomelic chondrodysplasia punctata are lethal genetic disorders caused by defects in **peroxisome** biogenesis. We report here a characterization of the **peroxisomal** matrix protein import capabilities of fibroblasts from 62 of these **peroxisome** biogenesis disorder patients representing all ten known complementation groups. Using an immunofluorescence microscopy assay, we identified three distinct **peroxisomal** protein import defects among these patients. Type-1 cells have a specific inability to import proteins containing the PTS1 **peroxisomal targeting** signal, type-2 cells have a specific defect in import of proteins containing the PTS2 signal, and type-3 cells exhibit a loss of, or reduction in, the import of both PTS1 and PTS2 proteins. Considering that the common cellular phenotype of Zellweger syndrome, neonatal adrenoleukodystrophy and infantile Refsum's disease has been proposed to be a complete defect in **peroxisomal** matrix protein import, the observation that 85% (40/47) of the type-3 cell lines imported a low but detectable amount of both PTS1 and PTS2 proteins was surprising. Furthermore, different cell lines with the type-3 defect exhibited a broad spectrum of different phenotypes; some showed a complete absence of matrix protein import while others contained 50-100 matrix protein-containing **peroxisomes** per cell. We also noted certain relationships between the import phenotypes and clinical diagnoses: both type-1 cell lines were from neonatal adrenoleukodystrophy patients, all 13 type-2 cell lines were from classical rhizomelic chondrodysplasia punctata patients, and the type-3 import defect was found in the vast majority of Zellweger syndrome (22/22), neonatal adrenoleukodytrophy (17/19), and infantile Refsum's disease (7/7) patients. Our finding that all type-1 cell lines were from the second complementation group (CG2), all 13 type-2 cell lines were from CG11, and that cells from the eight remaining complementation groups only exhibit the type-3 defect indicates that mutations in particular genes give rise to the different types of **peroxisomal** protein import defects. This hypothesis is further supported by correlations between certain complementation groups and particular type-3 subphenotypes: all patient cell lines belonging to CG3 and CG10 showed a complete absence of **peroxisomal** matrix protein import while those from CG6, CG7, and CG8 imported some **peroxisomal** matrix proteins. However, the fact that cell lines from within particular complementation groups (CG1, CG4) could have different matrix protein import characteristics suggests that allelic heterogeneity also plays an important role in generating different import phenotypes in certain patients.(ABSTRACT TRUNCATED AT 400 WORDS)

9/3,AB/9 (Item 9 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09499197 95354665 PMID: 7628448
Peroxisomal and mitochondrial carnitine acetyltransferases of **Saccharomyces** cerevisiae are encoded by a single gene.
Elgersma Y; van Roermund CW; Wanders RJ; Tabak HF
Department of Biochemistry, E.C. Slater Institute, Amsterdam, The Netherlands.
EMBO journal (ENGLAND) Jul 17 **1995**, 14 (14) p3472-9, ISSN

0261-4189 Journal Code: EMB
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Carnitine acetyltransferase (CAT) is present in mitochondria and **peroxisomes** of oleate-grown **Saccharomyces** cerevisiae. Both proteins are encoded by the same gene, YCAT, which encodes a protein with a mitochondrial **targeting** signal (MTS) at the N-terminus, and a **peroxisomal targeting** signal type 1 (PTS-1) at the C-terminus. Deletion of both motifs revealed the presence of an additional internal **targeting** sequence. Import of CAT via this internal signal was shown to be dependent on PAS10, a protein which is required for the import of PTS-1 containing proteins. An interaction of PAS10 with this internal **targeting** signal was demonstrated using the yeast two-hybrid technique. Expression of the YCAT gene behind a heterologous promoter resulted in loss of **peroxisomal targeting**, indicating that differential **targeting** is controlled at transcriptional or translational level. Determination of the 5'-ends of YCAT mRNAs revealed that YCAT transcripts initiating after the first AUG were present in oleate-grown cells. These transcripts were virtually absent in acetate- or glycerol-grown cells. We propose that in response to oleate, shorter transcripts are produced from which the **peroxisomal** form of CAT is translated, resulting in a CAT protein without a MTS, which can be **targeted** to **peroxisomes**.

9/3,AB/10 (Item 10 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09493567 94245796 PMID: 7910611
Differential protein import deficiencies in human **peroxisome** assembly disorders.
Motley A; Hettema E; Distel B; Tabak H
Department of Biochemistry, E. C. Slater Institute, University of Amsterdam, The Netherlands.
Journal of cell biology (UNITED STATES) May **1994**, 125 (4) p755-67, ISSN 0021-9525 Journal Code: HMV
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

Two **peroxisome targeting** signals (PTSs) for matrix proteins have been well defined to date. PTS1 comprises a COOH-terminal tripeptide, SKL, and has been found in several matrix proteins, whereas PTS2 has been found only in **peroxisomal** thiolase and is contained within an NH2-terminal cleavable presequence. We have investigated the functional integrity of the import routes for PTS1 and PTS2 in fibroblasts from patients suffering from **peroxisome** assembly disorders. Three of the five complementation groups tested showed a general loss of PTS1 and PTS2 import. Two complementation groups showed a differential loss of **peroxisomal** protein import: group I cells were able to import a PTS1- but not a PTS2- containing reporter protein into their **peroxisomes**, and group IV cells were able to import the PTS2 but not the PTS1 reporter into aberrant, **peroxisomal** ghostlike structures. The observation that the PTS2 import pathway is intact only in group IV cells is supported by the protection of endogenous thiolase from protease degradation in group IV cells and its sensitivity in the remaining complementation groups, including the partialized disorder of group I. The functionality of the PTS2 import pathway and colocalization of endogenous thiolase with the **peroxisomal** membranes in group IV cells was substantiated further using immunofluorescence, subcellular fractionation, and immunoelectron microscopy. The phenotypes of group I and IV cells provide the first evidence for differential import deficiencies in higher eukaryotes. These phenotypes are analogous to those found in **Saccharomyces** cerevisiae **peroxisome** assembly mutants.

9/3,AB/11 (Item 11 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09489668 95076712 PMID: 7985420
The **peroxisomal targeting** signal of 3-oxoacyl-CoA thiolase from **Saccharomyces** cerevisiae.
Erdmann R
Laboratory of Cell Biology, Howard Hughes Medical Institute, Rockefeller University, New York, NY 10021.
Yeast (ENGLAND) Jul **1994**, 10 (7) p935-44, ISSN 0749-503X Journal Code: YEA
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
All **peroxisomal** 3-oxoacyl-CoA thiolases identified so far do not contain the previously identified tripeptide **peroxisomal targeting** signal at their carboxy-termini. For the two rat thiolases it was shown that their **peroxisomal targeting** signals are localized within the amino-terminal region of the proteins and are cleaved upon import. This report demonstrates that the N-terminal region of the **peroxisomal** 3-oxoacyl-CoA thiolase from **Saccharomyces** cerevisiae is essential for its **peroxisomal targeting**, and that the N-terminal 16 amino acids of yeast thiolase are sufficient to **target** the otherwise cytosolic small subunit of ribulose-1,5-bisphosphate carboxylase to **peroxisomes** for import.

9/3,AB/12 (Item 12 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09489511 95071428 PMID: 7980572
The tetratricopeptide repeat-domain of the PAS10 protein of **Saccharomyces** cerevisiae is essential for binding the **peroxisomal targeting** signal-SKL.
Brocard C; Kragler F; Simon MM; Schuster T; Hartig A
Institut fur Biochemie und Molekulare Zellbiologie der Universitat Wien, Austria.
Biochemical and biophysical research communications (UNITED STATES) Nov 15 **1994**, 204 (3) p1016-22, ISSN 0006-291X Journal Code: 9Y8
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The PAS10 gene was found in a two-hybrid screen for the isolation of genes encoding proteins which interact with the C-terminal **peroxisomal targeting** signal -SKL. The PAS10 protein is known to be involved in import of proteins into **peroxisomes** and to contain a tetratricopeptide repeat (TPR) domain. All TPR-containing proteins involved in diverse processes like mitosis or RNA-synthesis share the ability to interact with other proteins. Here we show that the PAS10 protein interacts in vivo with the C-terminal **peroxisomal targeting** signal. The part essential for this interaction contains the complete tetratricopeptide repeat domain.

9/3,AB/13 (Item 13 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09488985 95050986 PMID: 7962087
An oligomeric protein is imported into **peroxisomes** in vivo.
McNew JA; Goodman JM
Department of Pharmacology, University of Texas Southwestern Medical Center, Dallas 75235-9041.
Journal of cell biology (UNITED STATES) Dec **1994**, 127 (5) p1245-57, ISSN 0021-9525 Journal Code: HMV

Contract/Grant No.: GM 31859, GM, NIGMS; T32 GM-07062, GM, NIGMS
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

The mechanism of translocation of **peroxisomal** proteins from the cytoplasm into the matrix is largely unknown. We have been studying this problem in yeast. We show that the **peroxisomal targeting** sequences SKL or AKL, with or without a spacer of nine glycines (G9), are sufficient to **target** chloramphenicol acetyltransferase (CAT) to **peroxisomes** of **Saccharomyces** cerevisiae in vivo. The mature form of CAT is a homotrimer, and complete trimerization of CAT was found to occur within a few minutes of synthesis. In contrast, import, measured by immunoelectron microscopy and organellar fractionation, occurred over several hours. To confirm that import of preassembled CAT trimers was occurring, we co-expressed CAT-G9-AKL with CAT lacking a **peroxisomal targeting** sequence but containing a hemagglutinin-derived epitope tag (HA-CAT). We found that HA-CAT was not imported unless it was co-expressed with CAT-G9-AKL. Both proteins were released from the organelles under mild conditions (pH 8.5) that released other matrix proteins, indicating that import had occurred. These results strongly suggested that HA-CAT was imported as a heterotrimer with CAT-G9-AKL. The process of oligomeric import also occurs in animal cells. When HA-CAT was co-expressed with CAT-G9-AKL in CV-1 cells, HA-CAT co-localized with **peroxisomes** but was cytoplasmic when expressed alone. It is not clear whether the import of globular proteins into **peroxisomes** occurs through **peroxisomal** membrane pores or involves membrane internalization. Both possibilities are discussed.

9/3,AB/14 (Item 14 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09483695 94256936 PMID: 8198533

Targeting of passenger protein domains to multiple intracellular membranes.

Janiak F; Glover JR; Leber B; Rachubinski RA; Andrews DW

Department of Biochemistry, McMaster University, Hamilton, Ontario, Canada.

Biochemical journal (ENGLAND) May 15 **1994**, 300 (Pt 1) p191-9, ISSN 0264-6021 Journal Code: 9YO

Languages: ENGLISH
Document type: Journal Article
Record type: Completed

The role of passenger domains in protein **targeting** was examined by fusing previously characterized **targeting** motifs to different protein sequences. To compare the **targeting** requirements for a variety of subcellular compartments, **targeting** of the fusion proteins was examined for endoplasmic reticulum, mitochondria and **peroxisomes** in vitro and in yeast. Although most passenger domains were only partially passive to translocation, motif-dependent **targeting** via motifs positioned at either end of one passenger domain (gPA) was demonstrated for all of the subcellular compartments tested. The data presented extend earlier suggestions that translocation competence is an intrinsic property of the passenger protein. However, the properties that determine protein **targeting** are not mutually exclusive for the compartments tested. Therefore, although the primary determinant of specificity is the **targeting** motif, our results suggest that translocation competence of the **targeted** protein augments the fidelity of transport.

9/3,AB/15 (Item 15 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09445645 98036045 PMID: 9370278

Isocitrate lyase localisation in **Saccharomyces** cerevisiae cells.

Chaves RS; Herrero P; Ordiz I; Angeles del Brio M; Moreno F
Departamento de Bioquimica y Biologia Molecular, Instituto Universitario de Biotecnologia de Asturias, Universidad de Oviedo, Spain.
Gene (NETHERLANDS) Oct 1 **1997**, 198 (1-2) p165-9, ISSN 0378-1119 Journal Code: FOP
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The isocitrate lyase from **Saccharomyces** cerevisiae was only located in the cell cytoplasm. This protein was found not to be associated with cell organelles, even under growth conditions that induce **peroxisome** proliferation. This conclusion is supported by experiments carried out by damaging the protoplast plasma membrane with DEAE-dextran, by differential centrifugation of osmotically lysed protoplast and by using the green fluorescent protein (GFP) of Aequorea victoria as a reporter fusion tag to localise the subcellular compartment to which isocitrate lyase is **targeted**.

9/3,AB/16 (Item 16 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09419684 97480732 PMID: 9339377
Genomic organization and molecular characterization of a gene encoding HsPXF, a human **peroxisomal** farnesylated protein.
Kammerer S; Arnold N; Gutensohn W; Mewes HW; Kunau WH; Hofler G; Roscher AA; Braun A
Institute of Anthropology and Human Genetics, University of Munich, Germany.
Genomics (UNITED STATES) Oct 1 **1997**, 45 (1) p200-10, ISSN 0888-7543 Journal Code: GEN
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
A protein modification essential for the cellular sorting of many biologically relevant proteins is the covalent attachment of prenyl lipids by specific transferases. Isoprenylation is known to render protein domains hydrophobic, thereby facilitating the interaction with lipid bilayers and/or membrane proteins. The **target** for the modification with farnesyl groups is the COOH-terminal sequence CaaX. Among the variety of farnesylated proteins the only one reported so far to be located to **peroxisomes** is the 37-kDa **peroxisomal** farnesylated hamster protein PxF. Recently we published data on the cDNA of the human gene HK33 (A. Braun et al., 1994, Gene 146: 291-295), which was revealed to be the human ortholog of PxF and was consequently renamed HsPXF. The genomic structure, molecular characterization, and evolutionary conservation of HsPXF are described herein. The exact location of the gene was defined as chromosome 1q22. The gene spans a region of approximately 9 kb, containing eight exons and seven introns. The 5' upstream region showed two potential Sp1-binding sites and an Alu repetitive sequence. Luciferase reporter activating capacity confirmed the presumed promoter activity of this region. On the transcriptional level, we detected four splice variants originating either from exon skipping or from alternative splicing events. For the HsPXF protein, a carboxyterminal farnesylation at cysteine residues was demonstrated. Through the use of HsPXF-specific antibodies, the protein was shown to be attached to the outer surface of **peroxisomes**. This localization together with the similarity to a **peroxisomal** assembly protein from **Saccharomyces** cerevisiae suggests HsPXF is involved in the process of **peroxisomal** biogenesis or assembly.

9/3,AB/17 (Item 17 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09417955 97459935 PMID: 9312008

Pex14p is a member of the protein linkage map of Pex5p.
Brocard C; Lametschwandtner G; Koudelka R; Hartig A
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EMBO journal (ENGLAND) Sep 15 **1997**, 16 (18) p5491-500, ISSN 0261-4189 Journal Code: EMB
Languages: ENGLISH
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To identify members of the translocation machinery for **peroxisomal** proteins, we made use of the two-hybrid system to establish a protein linkage map centered around Pex5p from **Saccharomyces** cerevisiae, the receptor for the C-terminal **peroxisomal targeting** signal (PTS1). Among the five interaction partners identified, Pex14p was found to be induced under conditions allowing **peroxisome** proliferation. Deletion of the corresponding gene resulted in the inability of yeast cells to grow on oleate as well as the absence of **peroxisomal** structures. The PEX14 gene product of approximately 38 kDa was biochemically and ultrastructurally demonstrated to be a **peroxisomal** membrane protein, despite the lack of a membrane-spanning domain. This protein was shown to interact with itself, with Pex13p and with both PTS receptors, Pex5p and Pex7p, indicating a central function for the import of **peroxisomal** matrix proteins, either as a docking protein or as a releasing factor at the organellar membrane.

9/3,AB/18 (Item 18 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09337208 97330864 PMID: 9187299
Nucleotide sequence of human alkyl-dihydroxyacetonephosphate synthase cDNA reveals the presence of a **peroxisomal targeting** signal 2.
de Vet EC; van den Broek BT; van den Bosch H
Department Biochemistry of Lipids, Centre for Biomembranes and Lipid Enzymology, Utrecht University, The Netherlands.
Biochimica et biophysica acta (NETHERLANDS) May 17 **1997**, 1346 (1) p25-9, ISSN 0006-3002 Journal Code: A0W
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Two overlapping clones were isolated from a human liver cDNA library in lambda gt11 that coded for human alkyl-dihydroxyacetonephosphate synthase using guinea pig and PCR-derived human cDNA probes. The open reading frame encodes a protein of 658 amino acids that shows a homology of 92% with the guinea pig homolog and a similarity of 98%. The **peroxisomal targeting** signal 2 that was recently identified in the presequence of the guinea pig enzyme appeared to be completely preserved in the human enzyme. Supportive confirmation for parts of the sequence of the mature protein was obtained from the Expressed Sequence Tags database of the National Center for Biotechnology Information. This database contained nine cDNA sequences, derived from seven independent clones, that correspond exactly to parts of the cDNA of human alkyl-dihydroxyacetonephosphate synthase. One of these clones most likely represents a not fully processed RNA with a putative intron containing an Alu sequence. An unexpected homology with D-lactate dehydrogenase (cytochrome C) precursor from **Saccharomyces** cerevisiae and with glycolate oxidase subunit D from Escherichia coli was also revealed.

9/3,AB/19 (Item 19 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09287818 97245715 PMID: 9090383
Rhizomelic chondrodysplasia punctata is caused by deficiency of human

PEX7, a homologue of the yeast PTS2 receptor.
Purdue PE; Zhang JW; Skoneczny M; Lazarow PB
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Nature genetics (UNITED STATES) Apr **1997**, 15 (4) p381-4, ISSN 1061-4036 Journal Code: BRO
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Languages: ENGLISH
Document type: Journal Article
Record type: Completed
The rhizomelic form of chondrodysplasia punctata (RCDP) is an autosomal recessive disease of **peroxisome** biogenesis characterized by deficiencies in several **peroxisomal** proteins, including the **peroxisomal** enzymes of plasmalogen biosynthesis and **peroxisomal** 3-ketoacyl thiolase. In cultured fibroblasts from patients with this disorder, both the **peroxisomal targeting** and proteolytic removal of the amino-terminal type 2 **peroxisomal targeting** sequence (PTS2) of thiolase are defective, whereas the biogenesis of proteins **targeted** by carboxyterminal type 1 **peroxisomal targeting** sequences (PTS1) is unimpaired. We have previously isolated a **Saccharomyces** cerevisiae **peroxisomal** biogenesis mutant, pex7 (formerly peb1/pas7), which demonstrates a striking similarity to the cellular phenotype of RCDP fibroblasts in that PTS1 **targeting** is functional, but the **peroxisomal** packaging of PTS2 **targeted** thiolase is lacking. Complementation of this mutant has led to the identification of the protein ScPex7p, a PTS2 receptor. In this paper we report cloning of the human orthologue of ScPEX7, and demonstrate that this is the defective gene in RCDP. We show that expression of human PEX7 in RCDP cells rescues PTS2 **targeting** and restores some activity of dihydroxyacetone phosphate acyltransferase (DHAP-AT), a **peroxisomal** enzyme of plasmalogen biosynthesis, and we identify the mutations responsible for loss of function of PEX7 in a compound heterozygote RCDP patient. These results imply that several **peroxisomal** proteins are **targeted** by PTS2 signals and that the various biochemical and clinical defects in RCDP result from a defect in the receptor for this class of PTS.

9/3,AB/20 (Item 20 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09287817 97245714 PMID: 9090382
Rhizomelic chondrodysplasia punctata is a **peroxisomal** protein **targeting** disease caused by a non-functional PTS2 receptor.
Motley AM; Hettema EH; Hogenhout EM; Brites P; ten Asbroek AL; Wijburg FA ; Baas F; Heijmans HS; Tabak HF; Wanders RJ; Distel B
Department of Biochemistry, University of Amsterdam, The Netherlands.
Nature genetics (UNITED STATES) Apr **1997**, 15 (4) p377-80, ISSN 1061-4036 Journal Code: BRO
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Rhizomelic chondrodysplasia punctata (RCDP) is an autosomal recessive disease characterized clinically by a disproportionately short stature primarily affecting the proximal parts of the extremities, typical dysmorphic facial appearance, congenital contractures and severe growth and mental retardation. Although some patients have single enzyme deficiencies, the majority of RCDP patients (86%) belong to a single complementation group (CG11, also known as complementation group I, Amsterdam nomenclature). Cells from CG11 show a tetrad of biochemical abnormalities: a deficiency of i) dihydroxyacetonephosphate acyltransferase, ii) alkyldihydroxyacetonephosphate synthase, iii) phytanic acid alpha-oxidation and iv) inability to import **peroxisomal** thiolase. These deficiencies indicate involvement of a component required for correct **targeting** of these **peroxisomal** proteins. Deficiencies in **peroxisomal**

targeting are also found in **Saccharomyces** cerevisiae pex5 and pex7 mutants, which show differential protein import deficiencies corresponding to two **peroxisomal targeting** sequences (PTS1 and PTS2). These mutants lack their PTS1 and PTS2 receptors, respectively. Like S. cerevisiae pex cells, RCDP cells from CG11 cannot import a PTS2 reporter protein. Here we report the cloning of PEX7 encoding the human PTS2 receptor, based on its similarity to two yeast orthologues. All RCDP patients from CG11 with detectable PEX7 mRNA were found to contain mutations in PEX7. A mutation resulting in C-terminal truncation of PEX7 cosegregates with the disease and expression of PEX7 in RCDP fibroblasts from CG11 rescues the PTS2 protein import deficiency. These findings prove